

IN THE CLAIMS

1. (currently canceled)

2 (withdrawn) The substrate according to claim 1 wherein said clostridial neurotoxin is botulinum neurotoxin serotype A. (currently canceled)

3 (withdrawn). The substrate according to claim 2 wherein said substrate is a peptide identified in SEQ ID NO:1 or SEQ ID NO:2.

4. (currently canceled)

5. (currently amended) A botulinum neurotoxin serotype B or tetanus toxin [The] substrate [according to claim 4] containing a signal moiety on one side of the cleavage site that produces a signal and a moiety that quenches the magnitude of said signal on the other side of the cleavage site such that when the substrate is cleaved, an increase in signal is produced and wherein said substrate is a peptide identified in SEQ ID NO:3 or SEQ ID NO:4.

6. (withdrawn) The substrate according to claim 1 wherein said clostridial neurotoxin is botulinum neurotoxin serotype D or botulinum neurotoxin serotype F.

7. (withdrawn) The substrate of to claim 6 wherein said substrate is chosen from the group consisting of a peptide identified in SEQ ID NO:5, SEQ ID NO:6, and SEQ ID NO:7.

8. (withdrawn) A method for detecting the presence of clostridial neurotoxin proteolytic acitivity in a sample said method comprising

mixing the sample with a peptide substrate according to claim 1, and

detecting an increase in signal produced from proteolytic cleavage of said substrate.

9. (withdrawn) A method for measuring concentration of neurotoxin in a sample, comprising

mixing the sample with a peptide substrate according to claim 1,

measuring an increase in signal with time produced from proteolytic cleavage of said substrate and,

determining the concentration of said neurotoxin by correlation to a standard.

10. (withdrawn) The method according to claim 9 wherein said neurotoxin is botulinum neurotoxin serotype A.

11. (withdrawn) The method according to claim 10 wherein said peptide substrate is a peptide identified in SEQ ID NO:1 or SEQ ID NO:2.

12. (withdrawn) The method according to claim 9 wherein said neurotoxin is botulinum neurotoxin serotype B or tetanus toxin.

13. (withdrawn - currently amended) [The] A method for detecting the presence of botulinum neurotoxin B or tetanus

toxin proteolytic activity in a sample, said method comprising:

mixing the sample with one or both [according to claim 12 wherein said] peptide substrates [is a peptide] identified in SEQ ID NO:3 or SEQ ID NO:4, and

detecting an increase in signal produced from proteolytic cleavage of said substrate.

14. (withdrawn) The method according to claim 9 wherein said neurotoxin is botulinum neurotoxin serotype D or F.

15. (withdrawn) The method according to claim 14 wherein said peptide substrate is chosen from the group consisting of a peptide identified in SEQ ID NO:5, SEQ ID NO:6, and SEQ ID NO:7.

16. (currently amended) A kit for determining the concentration of [a clostridial] botulinum neurotoxin serotype B or tetanus toxin in a sample, the kit containing in close confinement,

(i) one or [more] both peptide substrates according to claim [1] 5 cleavable by said [clostridial] botulinum neurotoxin or said tetanus toxin; and

(ii) said [clostridial] botulinum neurotoxin or said tetanus toxin standard.

17. (withdrawn) The kit according to claim 16 wherein said clostridial neurotoxin is botulinum neurotoxin serotype A and the peptide substrate is one or both of the peptides identified in SEQ ID NO:1 and SEQ ID NO:2.

18. (currently canceled)

19. (withdrawn) The kit according to claim 16 wherein said clostridial neurotoxin is botulinum neurotoxin serotype D or F and the peptide substrate is one or more of the peptides identified in SEQ ID NO:5, SEQ ID NO:6, and SEQ ID NO:7.

20. (currently canceled) A botulinum neurotoxin substrate comprising any peptide or protein that can serve as a substrate for the proteolytic activity of any clostridial neurotoxin, said protein or peptide having been modified so that it can be attached on one side of the proteolytic cleavage site to a solid material.

21. (withdrawn) The substrate according to claim 20 wherein said clostridial neurotoxin is botulinum neurotoxin serotype A.

22. (withdrawn) The substrate according to claim 21 wherein said substrate is a peptide identified in SEQ ID NO:8 or SEQ ID NO:11.

23. (currently amended) [The] A botulinum neurotoxin serotype B or a tetanus toxin substrate [according to claim 20] comprising a peptide or protein which may be immobilized to a solid material and which contains a moiety that produces a measurable signal such that when the substrate is cleaved, the signal is released, and wherein said substrate is a peptide identified in SEQ ID NO:9.

24. (currently canceled)

25. (withdrawn) The substrate according to claim 20 wherein said clostridial neurotoxin is botulinum neurotoxin serotype D or serotype F.

26. (withdrawn) The substrate of claim 25 wherein said substrate is a peptide identified in SEQ ID NO:10.

27. (withdrawn) The substrate of claim 20 wherein said botulinum neurotoxin is botulinum neurotoxin serotype E.

28. (withdrawn) The substrate of claim 27 wherein said substrate is a peptide identified in SEQ ID NO:11 and 12.

29. (withdrawn) A method for detecting the presence of clostridial neurotoxin proteolytic activity in a sample said method comprising

mixing the sample with a peptide substrate according to claim 20, and

detecting an increase in signal produced from proteolytic cleavage of said substrate.

30. (withdrawn) A method for measuring concentration of neurotoxin in a sample, comprising

mixing the sample with a peptide substrate according to claim 20,

measuring an increase in signal with time produced from proteolytic cleavage of said substrate and,

determining the concentration of said neurotoxin by correlation to a standard.

31. (withdrawn) The method according to claim 30 wherein said neurotoxin is botulinum neurotoxin serotype A.

32. (withdrawn) The method according to claim 31 wherein said peptide substrate is a peptide identified in SEQ ID NO:8 or SEQ ID NO:11.

33. (withdrawn) The method according to claim 30 wherein said neurotoxin is botulinum neurotoxin serotype B or tetanus toxin.

34. (withdrawn - currently amended) [The] A method for measuring concentration of botulinum neurotoxin B or tetanus toxin in a sample, comprising

mixing the sample with a peptide substrate [according to claim 33 wherein said peptide substrate is a peptide] identified in SEQ ID NO:9,

measuring an increase in signal with time produced from proteolytic cleavage of said substrate and,

determining an increase the concentration of said neurotoxin by correlation to a standard.

35. (withdrawn) The method according to claim 30 wherein said neurotoxin is botulinum neurotoxin serotype D or F.

36. (withdrawn) The method according to claim 35 wherein said peptide substrate is a peptide identified in SEQ ID NO:10.

37. (withdrawn) The method according to claim 30 wherein said neurotoxin is botulinum neurotoxin serotype E.

38. (withdrawn) The method according to claim 37 wherein said peptide substrate is a peptide identified in SEQ ID NO:11 or SEQ ID NO:12.

39. (currently amended) A kit for determining the concentration of [a clostridial] botulinum neurotoxin serotype B or tetanus toxin in a sample, the kit containing in close confinement,

(i) [one or more] the peptide [substrates] substrate according to claim [20] 23 cleavable by said [clostridial] botulinum neurotoxin or said tetanus toxin; and

(ii) said [clostridial] botulinum neurotoxin or said tetanus toxin standard.

40. (withdrawn) The kit according to claim 39 wherein said clostridial neurotoxin is botulinum neurotoxin serotype A and the peptide substrate is one or both of the peptides identified in SEQ ID NO:8 and SEQ ID NO:11.

41. (currently canceled)

42. (withdrawn) The kit according to claim 39 wherein said clostridial neurotoxin is botulinum neurotoxin serotype D or F and the peptide substrate is a peptide identified in SEQ ID NO:10.

43. (withdrawn) The kit according to claim 39 wherein said clostridial neurotoxin is botulinum neurotoxin serotype E and the peptide substrate is one or more of the peptides identified in SEQ ID NO:11 or SEQ ID NO:12.

44. (withdrawn) A method for identifying inhibitors or enhancers of proteolytic activity of a clostridial neurotoxin comprising:

preincubating a neurotoxin with a test compound to make a neurotoxin-compound solution,

exposing said solution to a substrate of said neurotoxin according to claim 20,

measuring signal resulting from the proteolysis of said substrate by said neurotoxin, and

comparing said signal with controls,

wherein an increase in signal indicates a compound which enhances neurotoxin activity and a decrease in signal indicates a compound which inhibits said neurotoxin.

45. (withdrawn) The method according to claim 44 wherein said neurotoxin is botulinum neurotoxin serotype A and the

substrate is a peptide identified in SEQ ID NO:8 or SEQ ID NO:11.

46. (withdrawn - currently amended) [The] A method for identifying inhibitors or enhancers of proteolytic activity of [according to claim 44 wherein said neurotoxin is] botulinum neurotoxin B or tetanus [toxin] neurotoxin comprising:

preincubating the neurotoxin with a test compound to make a neurotoxin-compound solution

exposing said solution to [and the substrate is] a peptide identified in SEQ ID NO:9,

measuring signal resulting from the proteolysis of said substrate by said neurotoxin, and

comparing said signal with controls,

wherein an increase in signal indicates a compound which enhances neurotoxin activity and a decrease in signal indicates a compound which inhibits neurotoxin activity.

47. (withdrawn) The method according to claim 44 wherein said neurotoxin is botulinum neurotoxin serotype D or F and the substrate is a peptide identified in SEQ ID NO:10.

48. (withdrawn) The method according to claim 44 wherein said neurotoxin is botulinum neurotoxin serotype E and the substrate is one or more of the peptides identified in SEQ ID NO:11 or SEQ ID NO:12.

49. (withdrawn) A method for identifying a serotype of a clostridial neurotoxin in a sample suspected of containing a neurotoxin, the method comprising

incubating the sample with antibodies against each clostridial neurotoxin such that a neurotoxin is bound to its serotype-specific antibody,

removing unbound components,
adding activation solution such that clostridial
protease is activated,
adding solutions containing clostridial neurotoxin
peptide substrates according to claim 1 to said activated
protease,
detecting signal generated from proteolysis of said
substrate by said protease, wherein a signal above control
indicates presence of a neurotoxin, and
determining the serotype of the clostridial neurotoxin
by noting the specificity of the antibody.

50. (withdrawn) The method of claim 49 wherein the
antibodies are bound to a solid material.

51. (withdrawn) The method of claim 50 wherein the solid
material is a multiwell plate.

52. (withdrawn) The method of claim 51 wherein each well
contains an antibody specific for a different neurotoxin
serotype.

53. (withdrawn) The method of claim 49 wherein each
peptide substrate is labeled with a different signal.

54. (withdrawn) A kit for identifying a serotype of a
clostridial neurotoxin in a sample suspected of containing a
neurotoxin, comprising
serotype-specific antibodies
clostridial neurotoxin standards, and
peptide substrates according to claim 1.